A NOTE ON THE EFFECT OF ALCOHOL-BENZENE EXTRACTIVES ON JUVENILE WOOD SPECIFIC GRAVITY IN RED PINE¹

Chen Hui Lee

Professor of Forest Genetics College of Natural Resources, University of Wisconsin Stevens Point, WI 54481

(Received April 1985)

ABSTRACT

A 24-millimeter-thick disk specimen was sampled from each of the three 10-year-old trees representing each of the 40 wind-pollinated red pine (*Pinus resinosa* Ait.) families. A $12 - \times 12 - \times 12$ -mm cube was cut from the bark side along the east cardinal direction. Smith's maximum moisture content method was used to determine unextracted and extracted specific gravities. Overall mean specific gravity for unextracted and extracted wood specimens was 0.351 and 0.309, a difference of 0.042 being statistically significant at the 1% level. The correlation between unextracted and extracted specific gravities was also statistically significant (r² = 0.444) at the 1% level. The larger experimental error variance was associated with the presence of alcohol-benzene extractives in the wood, and the need of a larger sample size to obtain the identical degrees of precision was suggested when working with unextracted wood specimens.

Keywords: Red pine, statistical precision, sample size, specific gravity, extractives.

INTRODUCTION

Nonstructural constituents such as resin acids, essential oils, fats and fatty acids, etc. found in plant cell are collectively called extractives. Their presence leads to an overestimate of wood specific gravity (Taras and Saucier 1967; Posey and Robinson 1969; Taylor 1974; Lai et al. 1980) and consequently affects the estimation of pulp yield per unit volume of dry wood material and wood mechanical properties.

Because specific gravity is under strong genetic control (Zobel 1961), the improvement of this important wood quality through selection breeding can be very effective. In any tree improvement program, we are confronted with an enormous number of measurements conducted on a periodic basis. It is imperative to find an alternative approach that saves measurement time and still yields useful genetic information (such as identifying or screening for top performing materials). Determination of the genetic variation in extracted specific gravity can be very tedious for tree improvement workers when large-sized circular-shaped disk specimens must be used for a specific research need. The present study was designed to analyze the magnitude of the variation in the alcohol-benzene extractives and their effect on the determination of wood specific gravity and the sampling efficiency in young red pine (*Pinus resinosa* Ait.).

¹ Study funded by the University of Wisconsin–Stevens Point Personnel Development Grant No. 5408.

Wood and Fiber Science, 18(3), 1986, pp. 376-381 © 1986 by the Society of Wood Science and Technology

MATERIALS AND METHODS

In 1970, a red pine seedling seed orchard was established in Wisconsin Rapids, Wisconsin, with 3-0 planting stock grown from seed collected from 310 wind-pollinated families in 46 natural stands throughout Wisconsin. It was machine-planted at 1.8×2.4 meter spacing in a replicated randomized complete block design. Each family was represented by a four-tree plot per replication. The orchard was thinned in 1980 and a 24-mm-thick disk specimen from a tree per plot was obtained from 0.2 meter above the ground. A $12- \times 12- \times 12$ -mm cube containing 2 to 3 outermost growth increments was removed at the bark side along the east cardinal direction, debarked, and oven-dried to arrest mold development. Forty families were selected from the original 310 families so that there were a total of 120 wood specimens (40 families $\times 1$ tree/family $\times 3$ replications) examined.

Smith's (1954) maximum moisture content method was used to determine both unextracted and extracted specific gravity. All wood specimens were then immersed in a mixture of ethanol: benzene (1:2 v/v) to remove the extractives from the wood specimens following the modified ASTM procedure outlined by Goggans (1962). The difference in the oven-dry weight between unextracted and extracted wood specimens was considered the amount of extractives and expressed on the extracted oven-dry weight basis.

An analysis of variance was performed on each of two sets of specific gravity data according to the following scheme. Mean family specific gravities based on 3 replications were used as items in the simple correlation analysis with 38 degrees of freedom. This was done to determine the degrees of association between extracted and unextracted specific gravities.

Source of variation	Degrees of freedom	Mean squares	F-test	Expected M.S.
Family	39	M ₁	M_1/M_3	$\sigma_{e}^{2} + 3\sigma_{F}^{2}$
Blocks	2	M_2		$\sigma_{\rm e}^2 + 40\sigma_{\rm R}^2$
Error	78	M_3		$\sigma_{\rm e}{}^2$
Total	119			

RESULTS AND DISCUSSION

Red pine was selected for two reasons: it is an important Lake States tree species and is known for its genetic uniformity in growth characteristics (Fowler et al. 1970) and wood specific gravity (Agar et al. 1983). The genetic uniformity permits a greater degree of precision to study and concentrate on the effect of alcoholbenzene extractives on the specific gravity determination.

Useful genetic information on wood properties can be obtained if there is consistency in the sampling of the study material. Comparable wood quality data were obtained for the genetic study purposes when all wood specimens were sampled from the identical position along the trunk and from the identical cardinal direction at the same trunk position (Lee 1974, 1976). As far as the consistency principle is enforced, Lee (1979) found that the use of branch-wood was even adequate to evaluate the genetic variation of wood quality in European black pine (*Pinus nigra* Arnold). It was further documented that inclusion in the working sample of a few growth increments taken at the bark side of the identical cardinal direction for the specific gravity study (Lee and Wahlgren 1979) and measurement of cell-wall morphology on all cells formed in a single growth increment laid during the same growing season located in the identical cardinal direction (Lee and Quirk 1983) for the study purpose of the sampling efficiency were statistically valid and acceptable. The within-tree variation (vertically along the trunk and transversely from the pith outward) in wood properties is far greater than the between-tree variation (Goggans 1961); however, this problem can be minimized by following the consistency principle in the sampling of the wood specimens.

There were no significant between-family differences in wood specific gravity (SG), moisture content (MC), and alcohol-benzene extractives (ABE) content (Table 1). The study material used in the present work probably included no mature wood.

Effect of extractive content

On the extracted oven-dry weight basis, mean alcohol-benzene extractive content was 10.6% for red pine. The effect of the extractives on the determination of specific gravity is substantial: mean extracted and unextracted specific gravities were 0.309 and 0.351, respectively. The difference was significant at the 1% level (F = 57.25 with 1/78 degrees of freedom). This reiterates that the effect of the extractives on the determination of wood specific gravity warrants a careful assessment whenever the specific gravity-related wood properties are to be evaluated and compared. Furthermore, mean extracted specific gravity obtained in the present study was comparable to a previous red pine study (Lee and Wahlgren 1979) using the 30-ring increment core sample taken from each of 15 sample trees from a southwestern Lower Michigan study plantation. In that study, mean extracted specific gravities based on 2 and 3 outermost growth rings were 0.321 and 0.319, respectively.

A mechanism for overestimating specific gravity was proposed by Taylor (1974). The removal of alcohol-benzene extractives causes dimensional (volumetric) changes in the wood blocks. An increase in the tangential as well as in the longitudinal dimension and a decrease in the radial dimension are sufficient enough to induce a decrease in specific gravity.

When the changes in specific gravity due to the presence of extractives are expressed as a percent of extracted specific gravity, red pine registered 13.59%. For Eastern white pine (*Pinus strobus* L.), similar changes were 9.04% for juvenile wood (growth rings 10 through 20 numbered from the pith outward) and 7.81% for mature wood (rings 20 through 30) (Thor 1965). The figures were still lower for the four major southern pines, ranging from 5.85% in slash pine (*P. elliottii* Engelm.) to 7.36% in longleaf pine (*P. palustris* Mill.) (Taras and Saucier 1967). This may be attributed to the differences in the physiology of tree growth.

It was not clear from the present study about the role of the age factor played in red pine. The trees used in this study were from a young, single age class (10 years in plantation age). As indicated in Table 2, there was a wide range in age

	Items	Wood specimens		
Wood properties		Unextracted	Extracted	
SG	mean	0.351	0.309	
	CV (%)	15.92	10.309	
	Sample size	69	31	
MC, %	mean	226	262	
	CV (%)	18.19	12.44	
	Sample size	91	42	
ABE	mean (%)		10.6	

TABLE 1. No between-family differences in wood properties.

CV = Coefficients of variation.

classes represented by the sample trees of different pine species and any direct comparison is probably unfeasible. Among the non-genetic factors, Posey and Robinson (1969) considered the age of the sample trees most important and influential on the amount of extractive content in the wood. They studied 480 shortleaf pine (P. echinata Mill.) trees ranging from 20 to 160 years in age from 48 locations in Oklahoma. The mean maximum overestimate due to extractives was 0.14 in actual specific gravity value in the juvenile wood (growth increments 1 through 10 numbered from the pith outward) and was only 0.08 in the mature wood (annual rings 11 through 20). The extractive content in the juvenile wood is more variable; thus the specific gravity may be overestimated from 0.02 to 0.53 in the juvenile wood. On the other hand, the extractive content remained relatively stable in the mature wood. In red pine, the size of the overestimate in real specific gravity was 0.042. This figure was much smaller in spite of the fact that the red pine sample trees were much younger in age and more variable in the extractive content than the shortleaf pine. Probably the genetic uniformity helped dilute the age effect.

Correlation between wood properties

The mean unextracted (independent variable) vs. extracted (dependent variable) specific gravity correlation was significant at the 1% level (y = 0.177 + 0.374x, where $r^2 = 0.444$). This was also the case with the southern pines ranging in the correlation coefficients of determination (= r^2) from 0.593 for loblolly pine (*P*.

Specific gravity No. of trees sampled Average age of trees Pine species Unextracted Extracted Sources Longleaf 143 50 0.530 Taras et al. 0.569 Loblolly 94 0.492 48 0.524 Ibid. Slash 123 30 0.543 0.513 Ibid. Shortleaf 140 62 0.524 0.491 Ibid. White 34 to 75 31 Young wood 0.398 0.365 Thor Old wood 0.414 0.384

TABLE 2. Differences in specific gravity due to the extractives in pine species.

taeda L.) with 92 degrees of freedom to 0.828 for slash pine with 121 degrees of freedom (Taras and Saucier 1967). These positive correlation coefficients simply imply that high unextracted specific gravity is associated with high extracted specific gravity. The strong correlation has a significant implication for tree improvement workers because the top performing genetic materials with the desirable wood quality can also be identified through the study of the unextracted wood specimens. If this approach is chosen, savings in the extraction effort can be substantial.

There was also a strong positive correlation between the amount of extractives in plant cells and unextracted specific gravity in this red pine study (r = 0.726). The strong relationship also held true for shortleaf pine (r = 0.75) reported by Posey and Robinson (1969). This means the overall effect of the extractive content can be predicted reasonably accurately from the unextracted wood specimens.

Sample size

The more fluctuating the amount of extractives is in the wood, the larger the sample size that is needed to achieve the identical degree of precision in the specific gravity study. From the data available for the southern pines with mean age ranging from 30 to 62 (Taras and Saucier 1967), I computed the sample size needed for both unextracted and extracted wood specimens. It was found necessary to increase the sample size by about 10 to 20% for the unextracted wood specimens. In red pine (10 years old), it requires up to 100% increase in the sample size or simply double the sample size (Table 1). It was not determined whether the contributing factors were the differences in the physiology of tree species or tree age or both.

The removal of alcohol-benzene extractives from the wood blocks may become tedious and expensive as the size of the wood specimens increases. This is particularly true when the investigator decides to select the circular-shaped disk specimens to meet the study need and when tens of thousands of wood samples must be handled as frequently is the case with tree improvement work. Doubling the sample size will certainly increase the workload; however, it may be warranted as an alternative in view of the tedious extraction process and the cost of alcohol and benzene. Besides, the determination of unextracted specific gravity is a reasonable alternative approach when the strong unextracted and extracted specific gravity relationship exists.

CONCLUSION

Mean specific gravity was 0.351 and 0.309, respectively, for unextracted and extracted red pine juvenile wood specimens. When expressed as a percent of extracted wood basis, specific gravity was overestimated by 13.59%. This was caused by the presence of 10.6% in the amount of alcohol-benzene extractives (expressed as a percent of the extracted oven-dry weight). There were no significant between-family differences in specific gravity, moisture content and the amount of the extracted specific gravities was established, and the size of the working sample should be doubled to achieve the identical degree of precision if we decide to determine wood specific gravity without the removal of the extractives.

REFERENCES

- AGAR, A., ET AL. 1983. Genetic gains from red pine seedling seed orchard. Proc. 28th Northeast Forest Tree Improve. Conf. Univ. New Hampshire, Durham, NH. Pp. 175–194.
- FOWLER, D. P., AND D. T. LESTER. 1970. Genetics of red pine. USDA Forest Service Res. Paper WO-8, 13 pp.
- GOGGANS, J. F. 1961. The interplay of environment and heredity as factors controlling wood properties in conifers with special emphasis on their effects on specific gravity. North Carolina Sta. Univ. School of For. Tech. Rep. No. 11. 56 pp.

-----. 1962. The correlation, variation and inheritance of wood properties in loblolly pine. North Carolina Sta. Univ. School of For. Tech. Rep. No. 14. 155 pp.

- LAI, Y. Z., ET AL. 1980. Wood and bark specific gravity determination as affected by water-soluble extractives loss. Wood Sci. 13(1):47-49.
- LEE, C. H. 1974. Geographic variation of growth and wood properties in eastern white pine. Proc. 21st Northeast For. Tree Improve. Conf. Pp. 36-41.

— 1976. Geographic variation of growth and wood properties in Japanese larch. Proc. 12th Lake States For. Tree Improve. Conf. Pp. 35–46.

——. 1979. Absence of growth-wood property correlation in 27 black pine seed sources. Wood Fiber 11(1):22–28.

-----, AND H. E. WAHLGREN. 1979. Specific gravity sampling with minimal tree damage—A study of red pine. Wood Sci. 11(4):241–245.

-----, AND J. T. QUIRK. 1983. Precision of measuring anatomical characteristics in jack pine. Wood Fiber Sci. 15(1):74–80.

- POSEY, C. E., AND D. W. ROBINSON. 1969. Extractives of shortleaf pine: An analysis of contributing factors and relationship. Tappi 52(1):110–115.
- SMITH, D. M. 1954. Maximum moisture content method for determining specific gravity of small wood samples. USDA Forest Service Forest Prod. Lab. Rep. No. 2014. 8 pp.
- TARAS, M. A., AND J. R. SAUCIER. 1967. Influence of extractives on specific gravity of southern pine. For. Prod. J. 17(9):97–99.
- TAYLOR, F. W. 1974. Effect of extraction on the volume determination and specific gravity of solid wood blocks. Wood Sci. 6(4):396-404.
- THOR, E. 1965. Variation in some wood properties of eastern white pine. Forest Sci. 11(4):451-455.
- ZOBEL, B. J. 1961. Inheritance of wood properties in conifers. Silvae Genet. 10:65-70. Frankfurt a.M., Germany.